



A new species of the genus *Ophiomyxa* (Echinodermata: Ophiuroidea: Ophiomyxidae) from the Mallorca Channel seamounts in the western Mediterranean

FRANCESC ORDINES^{1,3}, SERGIO RAMÍREZ-AMARO^{1,4*}, BELÉN CALERO², MARIA TERESA FARRIOLS^{1,4} & ENRIC MASSUTÍ^{1,5}

¹Centre Oceanogràfic de les Balears (COB-IEO), CSIC, Moll de Ponent s/n, 07015 Palma, Spain.

²Centro Oceanográfico de Murcia (COMU-IEO), CSIC, el Varadero, 1, 30740 Lo Pagan, Spain.

✉ belen.calero@ieo.csic.es; <https://orcid.org/0000-0002-4641-2022>

³✉ xisco.ordinas@ieo.csic.es; <https://orcid.org/0000-0002-2456-2214>

⁴✉ mt.farriols@ieo.csic.es; <https://orcid.org/0000-0002-7704-6504>

⁵✉ enric.massuti@ieo.csic.es; <https://orcid.org/0000-0002-9524-5873>

*Corresponding author: ✉ sergio.ramirez@ieo.csic.es; <https://orcid.org/0000-0002-0298-0749>

Abstract

A new species belonging to the ophiuroid genus *Ophiomyxa* is described from the Mallorca Channel seamount, in the western Mediterranean Sea. It can be distinguished from other *Ophiomyxa* species by the lack of interradial marginal plates, three arm spines, the presence of two thin, transparent and completely perforated dorsal arm plates on each arm segment, the separate heptagonal ventral arm plates, the disk integument full of transparent rounded scales with scattered perforated ossicles, and a characteristic coloration of the disk, which in the living specimen is brown with abundant scattered bright white spots. Molecular analyses based on cytochrome c Oxidase subunit I (DNA barcode) clearly support the assignment of the new species to *Ophiomyxa*. This discovery highlights the importance of the Mallorca Channel seamounts for the Mediterranean biodiversity conservation, as they seem to provide a suitable habitat for several invertebrate species, including recent descriptions of species and new Mediterranean records, which apparently have not established permanent populations along the closest continental margin.

Key words: Brittle star, Deep-sea, Balearic Islands, COI

Introduction

The Ophiuroidea (brittle stars and basket stars) is the most taxonomically diverse class of echinoderms, with at least 2133 described species (Stöhr *et al.* 2024). Ophiomyxidae is a taxonomically problematic family of ophiuroids that were originally placed in the order Phrynophiurida (Matsumoto 1915), then the order Ophiurida (Smith *et al.* 1995) and finally to the Ophiacanthida (O'Hara *et al.* 2018). This family is represented by 6 genera and 43 species. The most diverse genus is *Ophiomyxa* Müller & Troschel with 20 species (Stöhr *et al.* 2024). The great majority of ophiomyxid species were described before 1952, when *Ophiomyxa punctata* (A.H. Clark, 1952) was described. Since then, only two new ophiomyxid species have been described: *Ophiomyxa crinita* Franklin & O'Hara, 2008 and *Ophiocanis claviculata* Okanishi & Fujita, 2018.

Most of the *Ophiomyxa* species were described from the Pacific and Indian Oceans (up to 13), whereas seven are from the Atlantic Ocean, *Ophiomyxa brevicauda* Verrill, 1899, *Ophiomyxa flaccida* (Say, 1825), *Ophiomyxa stimpsonii* (Lyman, 1875), *Ophiomyxa tumida* Lyman, 1883, *Ophiomyxa serpentaria* Lyman, 1883 and *Ophiomyxa pentagona* (Lamarck, 1816). The first four are descriptions from the Caribbean (Say 1825; Lyman 1875, 1883; Verrill 1899), whereas *O. serpentaria* is distributed in the whole northeast Atlantic from the Azores to the Denmark Strait (Mortensen 1927). *Ophiomyxa pentagona* is distributed from the Cantabrian Sea to Cape Verde Archipelago and the northwest African coast (Tortonese 1965; Anadón 1977), and is currently considered the only known *Ophiomyxa* species from the Mediterranean. Along with *Ophiocanis forbesi* Heller, 1863, they are the only two Ophiomyxidae species reported from the Mediterranean Sea (Tortonese 1965).

The aim of the present work is to describe, based on morphological and molecular data, a new species of *Ophiomyxa* from Ses Olives seamount, in the Balearic Islands, western Mediterranean.

Material and Methods

Study area and sampling

The Balearic Islands (western Mediterranean) are located in the Balearic Promontory, a structural elevation of 348 km in length, 105 km wide and from 1000 to 2000 m high with respect to the surrounding basins. In the Mallorca Channel that separates the two subunits of this promontory (Mallorca-Menorca and Eivissa-Formentera), three seamounts are located (Figure 1): Ses Olives, with its summit at 225–290 m depth, and Ausias March and Emile Baudot with summits at 86–115 and 94–150 m depth, respectively (Massutí *et al.* 2022). While Ses Olives and Ausias March are of continental origin, Emile Baudot is of volcanic origin (Acosta *et al.* 2004). These seamounts are currently studied within the LIFE IP INTEMARES project, which aims to improve the scientific knowledge on their habitats and species, and human activities, in order to include them in the European network of marine Natura 2000 sites.

The surveys INTEMARES-A22B-0718, INTEMARES-A22B-1019 and INTEMARES-A22B-0720 were developed in 2018, 2019 and 2020, respectively, to study the seamounts of the Mallorca Channel. During these surveys, a total of 85 beam trawl samples (99–764 m depth) and 55 rock dredge samples (89–1191 m depth) were collected on sedimentary and rocky bottoms, respectively (Massutí *et al.* 2022).

The beam trawl used was a Jennings type with 2 and 0.5 m horizontal and vertical openings, respectively and a 5 mm mesh size cod-end. The efficiency of this beam trawl for sampling epibenthos has been estimated by Reiss *et al.* (2006). Sampling was conducted at 2 knots and between 5 and 15 min of effective sampling duration.

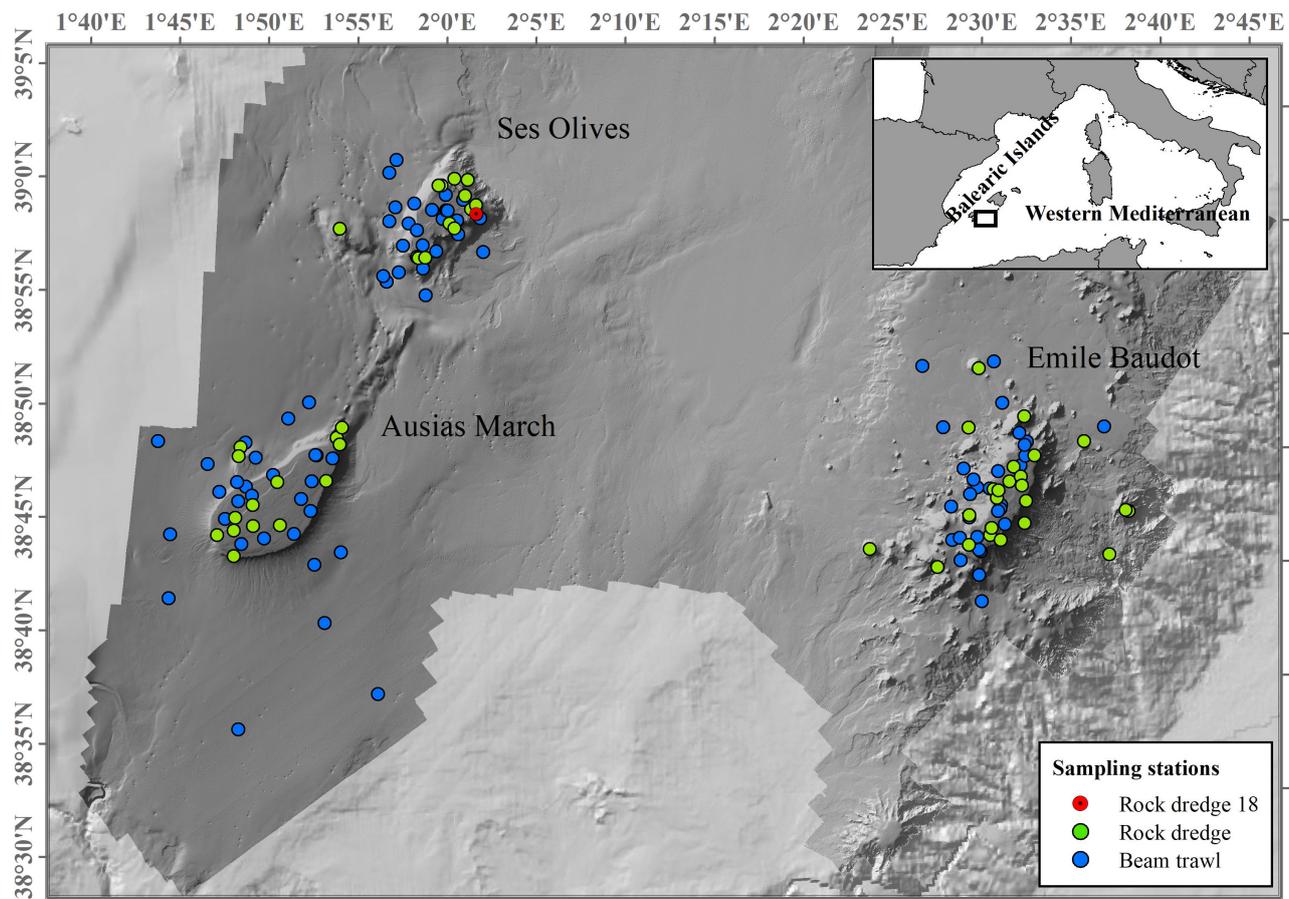


FIGURE 1. Map of the Mallorca Channel seamounts showing the sampling stations carried out using rock dredge and beam trawl during the INTEMARES surveys from 2018 to 2020. In red the sampling station where the described specimen was collected.

The rock dredge was composed of a metallic rectangular mouth with bevelled edges, equipped with a 1 cm mesh cod-end, protected by another net of 2 cm meshes and leather covers on bottom and top sides. It was trawled in an upward direction over the seafloor, collecting rock fragments, together with the associated flora and fauna. Sampling was conducted at 0.5–1 knots, with an effective duration from 5 to 10 min.

The specimen described here was collected during the INTEMARES-A22B-0718 survey, conducted in July 2018 on board the R/V *Ángeles Alvariño*. It was preserved in 96% ethanol. Later in the laboratory, two arm segments of a damaged arm were removed for genetic analyses. The specimen was analysed under a stereomicroscope Leica M165C equipped with a camera Leica MC170. Identification to genus was done according to Mortensen (1927) and Paterson (1985). The terminology applied for the morphological description follows Stöhr *et al.* (2012) and Hendlér (2018). The holotype of the new species has been deposited in the Marine Fauna Collection of the Centre Balear de Biodiversitat (<https://centrebaleardebiodiversitat.uib.cat>) with the identification reference number CBB-UIB 100485.

A solution 4% of sodium hypochlorite was used to remove the integument in comparative material.

Molecular analyses

Total DNA was extracted from a small piece of arm (~2 cm) of the collected specimen. DNA extraction was performed with the E.Z.N.A Tissue DNA kit (Omega Bio-tek) following the manufacturer's protocol. Polymerase chain reaction (PCR) was used to amplify the partial mitochondrial gene, cytochrome c Oxidase subunit I (COI; DNA barcode), with primers LCOech1aF1 (Corstorphine, 2010) and HCO2198 (Folmer *et al.* 1994). The PCR reactions were carried out in a final volume of 12.5 µL, containing 2 µL of template DNA, 0.5 µM of the primers, 6.25 µL of Supreme NZYTaQ 2x Green Master Mix (NZYTech), and ultrapure water up to 12.5 µL. The reaction mixture was incubated as follows: an initial denaturation step at 94 °C for 2 min, followed by 35 cycles of 94 °C for 30 s, 51°C for 1 min, 72 °C for 1 min, and a final extension step at 72 °C for 10 min. The PCR products were bidirectionally sequenced on an ABI 3730xl DNA Analyzer (Applied Biosystems, USA). DNA barcoding analyses were carried out by AllGenetics & Biology SL (www.allgenetics.eu).

Sequences were imported into BioEdit 7.0.5.2. (Hall 1999) and checked for quality and accuracy with nucleotide base assignment. The consensus DNA sequence was deposited in the GenBank database (<http://www.ncbi.nlm.nih.gov/genbank/>) under the accession number PP050560. To compare this resulting sequence with other species belonging the genus *Ophiomyxa*, we downloaded the COI sequences of the following species from GenBank: *O. anisacantha* H.L. Clark, 1911 (GenBank ID: AB758822), *O. australis* Lütken, 1869 (GenBank ID: KU895171), *O. bengalensis* Koehler, 1897 (GenBank ID: KU895173), *O. brevirima* H.L. Clark, 1949 (GenBank ID: KU895170), *O. crinita* (GenBank ID: KU895172), *O. flaccida* (GenBank ID: KU895175), *O. pentagona* (GenBank ID: KU895176), *O. serpentaria* (GenBank IDs: KF663496-98; ON341503, ON341511), *O. vivipara* Studer, 1876 (GenBank ID: KU895367). All these sequences have been published by Okanishi and Fujita (2013), Hugall *et al.* (2016) and Eichsteller *et al.* (2022). Three additional COI sequences were included: *O. brevicauda* (GenBank ID: PP156898), *O. stimpsonii* (GenBank ID: PP156900) and *O. tumida* (GenBank ID: PP156899). The final sequence matrix was aligned with ClustalW (Thompson *et al.* 1994).

Genetic distance (p-distance) and number of base differences between pairs of sequences were calculated with MEGA v.7.1 (Tamura *et al.*, 2013). Furthermore, a phylogenetic tree based on Bayesian Inference (BI) was reconstructed to understand the phylogenetic relationships of the new *Ophiomyxa* species. For this we used the GTR + I + G as the optimal substitution model, which was selected following the Bayesian Inference Criteria (BIC) using MEGA. BI was performed with MrBayes v.3.2.1 (Ronquist *et al.* 2012) conducting four independent MCMC runs (with four chains each) for 10 million generations, sampling every 2000 generations and discarding the first 25% of samples as burn-in. Convergence was assessed by effective sample size (ESS) calculation and was visualized using TRACER v.1.5. Two species were included as outgroups for the phylogenetic analysis: *Ophioderma longicaudum* (Bruzellius, 1805) (GenBank ID: KU895218), belonging to the family Ophiodermatidae, and *Ophiotreta spectabilis* (G.O. Sars, 1872) (GenBank ID: KU895367), belonging to the family Ophiocanthidae (Hugall *et al.* 2016).

Results

Systematics

Class OPHIUROIDEA Gray, 1840

Order OPHIACANTHIDA O'Hara, Hugall, Thuy, Stöhr & Martynov, 2017

Family OPHIOMYXIDAE Ljungman, 1867

Genus *Ophiomyxa* Müller & Troschel, 1840

Ophiomyxa xispa sp. nov. Ordines, Calero & Ramírez-Amaro

Figures 2 and 3

Type material. Holotype. CBB-UIB 100485, Ses Olives seamount, Mallorca Channel, Balearic Islands, western Mediterranean (38°57.38'N, 02°00.87'E) 235–263 m depth, rock dredge, R/V *Ángeles Alvariño*, Cruise INTEMARES-A22B_0718, St. 18, Francesc Ordines, 28 July 2018.

Comparative material. *Ophiomyxa serpentaria*: Senckenberg am Meer, German Centre for Marine Biodiversity Research in Hamburg (DZMB-HH) 61037 (one specimen), Reykjanes Ridge (60°14.86'N, 29°08.274'W) 718 m, ROV, R/V *Maria S. Merian*, Cruise IceAGE, St. 67-6, 13 July 2018. DZMB-HH 61728 and DZMB-HH 61729 (one specimen each), Reykjanes Ridge (60°14.25'N, 29°06.89'W) 644 m, ROV, R/V *Maria S. Merian*, Cruise IceAGE, St. 188-6, 1 August 2018. DZMB-HH 61728, Reykjanes Ridge (60°14.25'N, 29°06.89'W) 644 m, ROV, R/V *Maria S. Merian*, Cruise IceAGE, St. 188-6, 1 August 2018. Zoological collections of the Marine Zoology Laboratory at University of Vigo (LZM-UV) 7430 (three specimens), Morocco (35°15'31"N, 6°35'01"W), 874 m, bottom trawl, R/V *Vizconde de Eza*, Cruise Maroc-0411, St. MO-04, 14 November 2004. LZM-UV 16697 (one specimen), Western Sahara (26°15'48"N, 15°09'32"W), 843 m, bottom trawl, R/V *Vizconde de Eza*, Cruise Maroc-0511, St. MO179, 10 December 2005. LZM-UV 6702 (one specimen), Western Sahara (26°12'42"N, 15°27'44"W), 1311 m, bottom trawl, R/V *Vizconde de Eza*, Cruise Maroc-0511, St. MO184, 12 December 2005. LZM-UV 16725 (one specimen), Western Sahara (26°06'50"N, 15°26'49"W), 847 m, bottom trawl, R/V *Vizconde de Eza*, Cruise Maroc-0511, St. MO185, 12 December 2005. LZM-UV 7624 (one specimens), Western Sahara (25°44'01"N, 15°56'01"W), 805 m, bottom trawl, R/V *Vizconde de Eza*, Cruise Maroc-0611, St. MO281, 10 December 2006. LZM-UV 18008 (one specimens), Guinea Bissau (10°22'50"N, 17°19'08"W), 895 m, bottom trawl, R/V *Vizconde de Eza*, Cruise Bissau-0810, St. BS156, 14 November 2004. *Ophiomyxa pentagona*: Centre Balear de Biodiversitat-Universitat de les Illes Balears (CBB-UIB) 100026 (one specimen), south Formentera, Balearic Islands, western Mediterranean (38°35.21'N, 01°26.51'E) 54 m depth, bottom trawl, R/V *Miguel Oliver*, Cruise MEDITS_ES_2023, St. 263, Francesc Ordines, 19 August 2023. CBB-UIB 100487 (one specimen), Menorca Channel, Balearic Islands, western Mediterranean (39°40.58'N, 03°32.12'E) 60 m depth, beam trawl, R/V *Ángeles Alvariño*, Cruise CANAL0419, St. 20, Francesc Ordines, 28 March 2019.

Etymology. The species name *xispa* is the name of the first author's dog and also is the word in Catalan for spark, which alludes to the bright white spots on the brown background of the disk.

Diagnosis. The specimen is identified as belonging to the family Ophiomyxidae and genus *Ophiomyxa* due to the following combination of characters: disk, oral area, arms and arm spines covered by integument; radial shields very small (about 1/8 the disk diameter); broad infradental and oral papillae with serrated edge; second oral tentacle pore opens inside the mouth.

The specimen can be distinguished from the rest of *Ophiomyxa* species by the combination of the following characters: interradial marginal plates absent; arm spines 3; two thin, transparent and completely perforated dorsal arm plates on each segment, arranged longitudinally on the arm axis; ventral arm plates heptagonal, separated by lateral arm plates; disk integument full of transparent rounded scales and few scattered perforated ossicles; disk brown, covered with abundant scattered bright white spots.

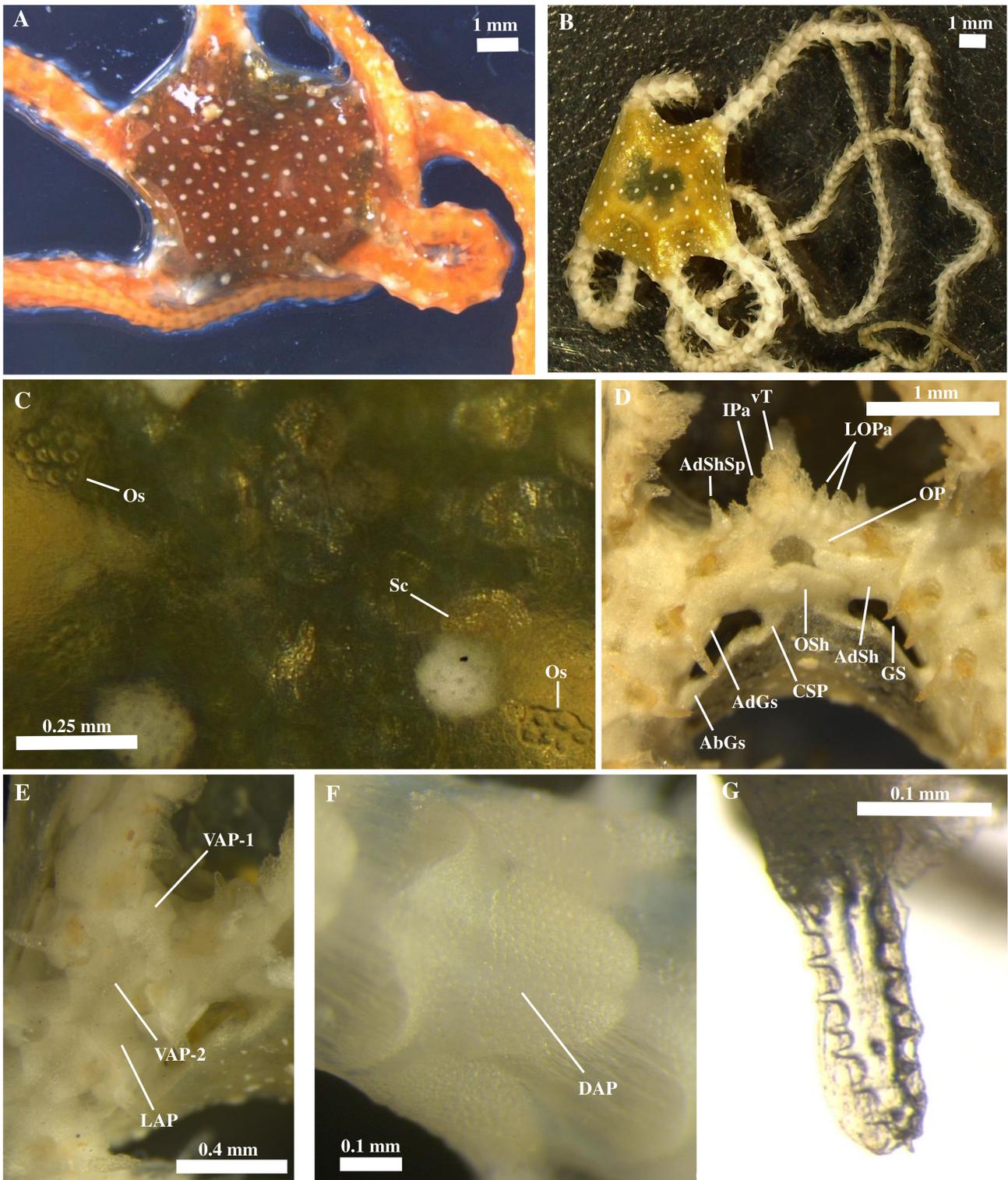


FIGURE 2. *Ophiomyxa xispa* sp. nov. A: dorsal view of the holotype alive. B: dorsal view of the holotype preserved in ethanol. C: Disk integument showing the scales and ossicles. D: Detail of the jaw and genital slits. E: detail of proximal arm segments. F: dorsal arm plate. G: detail of an arm spine. Abbreviations: AdSh, adoral shield; AdShSp, adoral shield spine; AdGs, adradial genital scale; AbGs, abradial genital scale; CSP, comma-shaped plate; DAP, dorsal arm plate; GS, genital slit; IPa, infradental papilla; LAP, lateral arm plate; LOPa, lateral oral papilla; OP, oral plate; Os, ossicle; Osh, oral shield; Sc, scales; VAP-1, first ventral arm plate; VAP-2, second ventral arm plate; vT, ventralmost tooth.

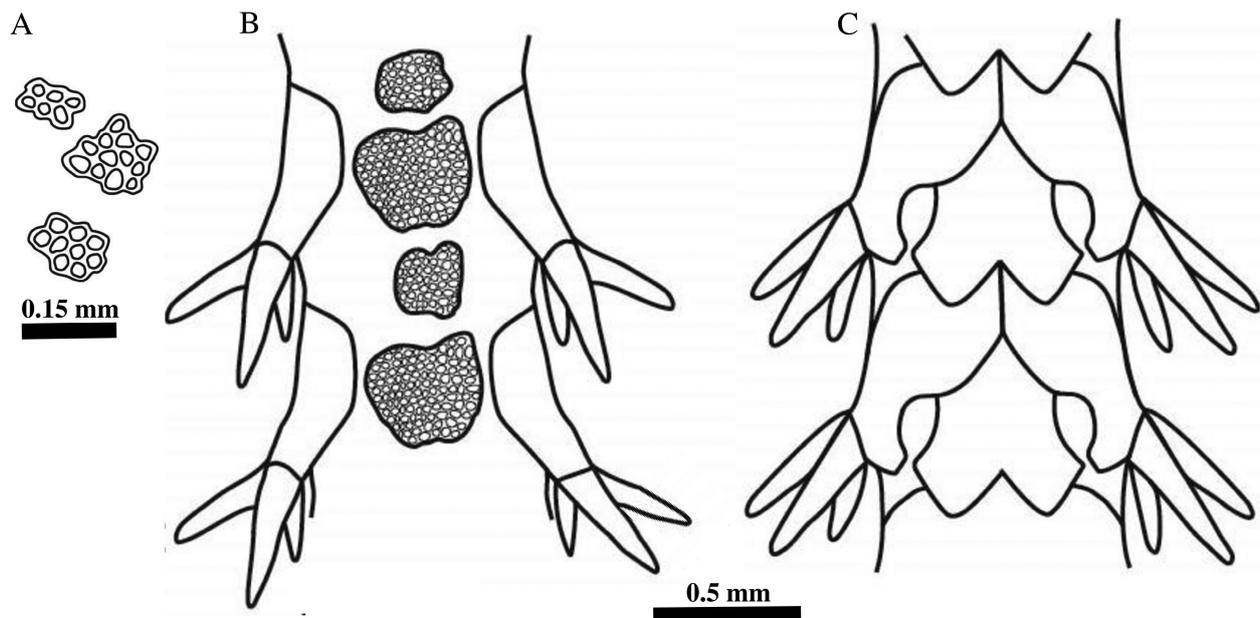


FIGURE 3. *Ophiomyxa xispa* sp. nov. ossicles from the disk integument (A), and dorsal (B) and ventral (C) arm plates. Drawing by N ria Ordines.

Description. Disk diameter 6.1 mm with slightly indented interradial margins. Disk covered with thin smooth integument, transparent so it is possible to distinguish the jaws from the dorsal side (Figure 2A, B). The integument is full of thin transparent rounded scales, their diameter ranging from 150 μ m to 210 μ m, as well as some scattered perforated ossicles of diameter 140 to 160 μ m and each bearing 5–12 perforations (Figures 2C and 3A). The integument also covers, but doesn't obscure, the oral and adoral shields, the base of the oral plates, the arms and the arm spines. Radial shields small, about 0.4 mm in length (1/8 the disk radius) and 0.2 mm in width, barely visible through integument. No series of interradial marginal plates, but 2–4 perforated plates appear at the base of the arms between the radial shields. Genital slits short, 0.7 mm long, barely reaching the end of the 2nd arm segment, ending 1 mm before the disk margin (Figure 2D). Adradial and abradial genital scales visible. Abradial genital scale almost in touch proximally with a comma-shaped plate partially overlapped by the oral shield (Figure 2D), the space in between filled with 1–3 small perforated plates.

Arms are approximately 38 mm long and 1 mm wide at the base. The ventral and lateral arm plates are barely visible through the integument in the live individual, whereas the dorsal arm plates are completely obscured. Once the individual was preserved in pure ethanol all arm plates were visible. The lateral arm plates are in contact ventrally from the second segment (Figure 2E), and are moderately extended dorsally. Dorsal arm plates are thin, delicate, transparent and completely perforated. They appear on the first two proximal segments as a continuous plate, with an ill-defined shape; from the third, each segment bears two plates, a proximal one nearly rounded but with very irregular margins, and a distal one larger also with irregular margins and nearly semicircular, the rounded edge oriented distally, with sides almost reaching the lateral arm plates (Figure 2F and 3B). Ventral arm plates heptagonal, with similar length and width, and separated by the lateral arm plates; on proximal segments they measure about 0.6 mm long and 0.55 mm wide; proximally they are sharply pointed whereas the distal end presents a deep notch (Figure 2E and 3C); all angles sharp. The 1st ventral plate with similar shape and proportions than the rest but with rounded angles (Figure 2E). Each lateral arm plate bears three spines positioned laterally, with the middle spine at the lateral midline, some segments bearing only two spines per side. The spines are serrated (Figure 2G) and all three very similar in length. Proximally and midway down the arm they measure 0.5–0.6 mm in length (about equal or slightly less than the length of a segment) and 0.1 mm in width, then decreasing gradually to around 0.2 mm in length (about half the length of a segment) and 0.05 mm in width, at the tip of the arms, where the spines are hooked at the last segments. There is only one spine on the arm segments within the disk. No tentacle scales. Second oral tentacle pore opens inside the mouth.

Oral shields nearly triangular with rounded lateral angles, about two times wider than long. Adoral shields wide, nearly trapezoid-shaped with the proximal edge in contact with the oral plates but not touching each other, leaving

a gap between adoral and oral shields and the oral plates (Figure 2D). Oral plates about 0.5 mm long and 0.2 mm wide. At each oral plate there are one infradental papillae and 1–3 lateral oral papillae. The infradental papillae is glassy, 1.5 higher than wide, with rounded serrated edge. The lateral oral papillae are smaller than the infradental, more triangular, about 3 times higher than wide, with serrated edge (Figure 2D). Dental plates bear three teeth, the ventralmost and the middle one similar to the infradental papillae (in two of the jaws not as serrated), the innermost about 2.5 higher than wide. Adoral shield spine pointed, well visible (Figure 2D).

Colour. Live specimen (Figure 2A): Dorsal surface of disk is brown, covered with abundant scattered bright white spots. The ventral disk surface is more transparent, appearing darker due to the stomach wall, and without or only with one white spot in each interradial space. Arms are orange dorsally and ventrally with tube feet a transparent white. Oral and adoral shields and oral plates and base of oral papillae orange. Preserved specimen (Figure 2B): the orange colour faded away completely and these parts appearing now white or transparent. The brown colour of the disk is now yellowish with white spots completely preserved and still standing out on the background.

Genetics and phylogeny. A fragment of 603 base pairs (bp) of the COI mitochondrial gene was sequenced. The nucleotide frequencies were T = 27.53, C = 27.03, A = 26.87 and G = 18.57. In general, high values of genetic distance as well as a high number of base pairs differences were detected between *Ophiomyxa* species, the average of both indices was of 17.6% and 106 pb differences. The new species *Ophiomyxa xispa* **sp. nov.** showed the closest genetic distance to *O. stimpsonii* (8.5% and 51 bp differences; Table 1). This genetic distance was among the closest interspecific distances observed of all the analyzed *Ophiomyxa* species. Closest distances were only found between *O. vivipara* and *O. serpentaria* (3.4% and 20 bp differences), *O. tumida* and *O. brevicauda* (5.6% and 34 bp differences) and *O. crinita* and *O. bengalensis* (7.3% and 44 bp differences). The phylogenetic reconstruction based on the COI fragment clearly separated *O. xispa* **sp. nov.** from the rest of species, and showed two main clades within the genus *Ophiomyxa*: one including *O. xispa* **sp. nov.**, *O. stimpsonii*, *O. neglecta*, *O. bengalensis*, and *O. crinita*, and another including *O. serpentaria*, *O. vivipara*, *O. anisacantha*, *O. brevirima*, *O. australis*, *O. flaccida*, *O. pentagona*, *O. brevicauda* and *O. tumida* (Figure 4). *Ophiomyxa stimpsonii* appeared as the phylogenetically closest species to *O. xispa* **sp. nov.** Likewise, the species with a closer interspecific distance showed a closer phylogenetic relationship (Figure 4).

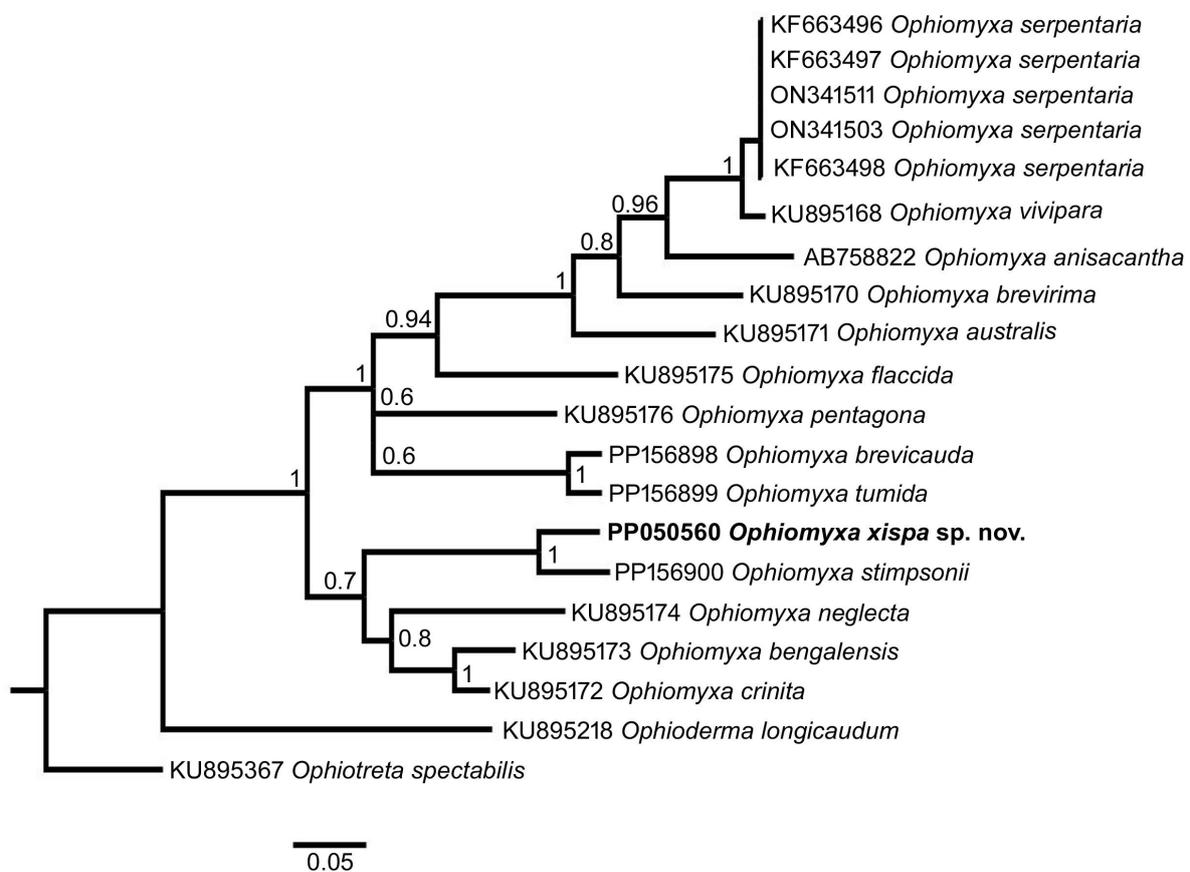


FIGURE 4. Phylogenetic tree based on Bayesian inference for a COI fragment for *Ophiomyxa* species. Posterior probabilities are indicated near the nodes. The new species is indicated in bold face.

TABLE 1. Mean genetic distances (%) and numbers of base differences for COI fragment of species pairs of *Ophiomyxa* species, below and above the diagonal, respectively.

| | Species | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
|----|--------------------------|------|------|------|------|------|------|------|------|------|------|------|------|------|-----|
| 1 | <i>O. xispa</i> sp. nov. | | 112 | 118 | 97 | 119 | 116 | 102 | 116 | 106 | 113 | 99 | 51 | 123 | 99 |
| 2 | <i>O. anisacantha</i> | 18.6 | | 95 | 113 | 115 | 79 | 114 | 123 | 117 | 121 | 75 | 120 | 116 | 74 |
| 3 | <i>O. australis</i> | 19.6 | 15.8 | | 126 | 115 | 85 | 126 | 94 | 118 | 107 | 93 | 114 | 120 | 92 |
| 4 | <i>O. bengalensis</i> | 16.1 | 18.7 | 20.9 | | 112 | 114 | 44 | 115 | 89 | 102 | 115 | 100 | 108 | 116 |
| 5 | <i>O. brevicauda</i> | 19.7 | 19.1 | 19.1 | 18.6 | | 112 | 105 | 106 | 101 | 103 | 125 | 122 | 34 | 123 |
| 6 | <i>O. brevirima</i> | 19.2 | 13.1 | 14.1 | 18.9 | 18.6 | | 118 | 112 | 110 | 111 | 86 | 118 | 113 | 87 |
| 7 | <i>O. crinita</i> | 16.9 | 18.9 | 20.9 | 7.3 | 17.4 | 19.6 | | 116 | 86 | 102 | 120 | 97 | 109 | 121 |
| 8 | <i>O. flaccida</i> | 19.2 | 20.4 | 15.6 | 19.1 | 17.6 | 18.6 | 19.2 | | 121 | 102 | 110 | 114 | 103 | 113 |
| 9 | <i>O. neglecta</i> | 17.6 | 19.4 | 19.6 | 14.8 | 16.7 | 18.2 | 14.3 | 20.1 | | 100 | 128 | 102 | 104 | 125 |
| 10 | <i>O. pentagona</i> | 18.7 | 20.1 | 17.7 | 16.9 | 17.1 | 18.4 | 16.9 | 16.9 | 16.6 | | 119 | 110 | 100 | 124 |
| 11 | <i>O. serpentaria</i> | 16.4 | 12.4 | 15.4 | 19.1 | 20.7 | 14.3 | 20.0 | 18.2 | 21.2 | 19.7 | | 111 | 116 | 20 |
| 12 | <i>O. stimpsonii</i> | 8.5 | 19.9 | 18.9 | 16.6 | 20.2 | 19.6 | 16.1 | 18.9 | 16.9 | 18.2 | 18.3 | | 130 | 112 |
| 13 | <i>O. tumida</i> | 20.4 | 19.2 | 19.9 | 17.9 | 5.6 | 18.7 | 18.1 | 17.1 | 17.2 | 16.6 | 19.3 | 21.6 | | 112 |
| 14 | <i>O. vivipara</i> | 16.4 | 12.3 | 15.3 | 19.2 | 20.4 | 14.4 | 20.1 | 18.7 | 20.7 | 20.6 | 3.4 | 18.6 | 18.6 | |

Ecology. The holotype was collected from a rocky bottom on the margin of Ses Olives seamount summit and the beginning of its upper flank. The rock dredge sample was mainly composed by coarse biogenic sand with mud and fragments of limestone crusts, with several sponges and brachiopods, as well as the bivalve *Asperarca nodulosa* (O.F. Müller, 1776) and the decapods crustaceans *Plesionika edwardsii* (Brandt, 1851) and *Plesionika narval* (Fabricius, 1787), all of them characteristic of rocky bottoms. The *Ophiomyxa* specimen was hidden inside a small crevice in one of the rock fragments (Figure 5). Despite a thorough sampling of the Mallorca Channel seamounts rocky and sedimentary bottoms (Figure 1), we have not recovered any other specimen of this species so far. However, high abundances of ophiuroids were detected in this area, particularly from sedimentary bottoms (Ordines *et al.* 2019; Massutí *et al.* 2022). So far, the species seems to be restricted to rocky bottoms.

Remarks. *Ophiomyxa xispa* sp. nov. is distinguished from *O. pentagona*, the only *Ophiomyxa* species recorded so far from the Mediterranean, by the number of arm spines (3 vs. 4–5, respectively), the shape of the ventral arm plates (as long as wide vs. 1.5–1.6 times wider than long, respectively; Figures 2E, 3C and 6A), and the shape and arrangement of dorsal arm plates: two plates, a proximal one nearly rounded, and a distal one larger nearly semicircular in *O. xispa* sp. nov. vs. two dorsal arm plates with triangular shape, about twice as wider as long, one on each side of the midline of the arm, where they are in contact; these plates are also in contact with the lateral arm plates (Figures 2F, 3B and 6B, C, D).

Our species morphologically resembles the subgroup of *Ophiomyxa* species characterised by the absence of a row of marginal interradial plates. The species with this character were previously distinguished as the genus *Ophiodera* Verrill, 1899, who differentiated it from *Ophiomyxa* mainly by the following characteristics (Verrill 1899; 1900): rudimentary marginal disk scales and absence of the disk-scales proximal to the radial shields, no upper arm plates, and, three or four arm-spines enclosed in cuticle. Verrill (1899) designated *O. serpentaria* as the type species for *Ophiodera*, and hence this genus is not currently accepted, as Mortensen (1927) considered it a synonym of *Ophiomyxa*. This synonymy was ignored by A. H. Clark when describing *O. punctata* as a member of *Ophiodera* (A.H. Clark 1952). Other species originally described as *Ophiodera* include *O. compacta* and *O. neglecta*, whereas *O. stimpsonii* was transferred to *Ophiodera* by Verrill (1900). The description of all these species fits within *Ophiodera*, with no interradial marginal scales or at least lacking the scales proximal to the radial shields as is the case for *O. stimpsonii* (Verrill 1900), no dorsal arm plates and 3–4 arm spines (rarely five in *O. punctata* and four proximally but five distally in *O. compacta*). *Ophiomyxa bengalensis*, described previously to the description of the genus *Ophiodera*, also shares the main differentiating characters of this genus and lacks interradial marginal plates, dorsal arm plates and has 3 arm spines (Koehler 1897). Finally, *O. anisacantha* H.L. Clark, 1911, originally described as *Ophiomyxa*, and *O. crinita*, have no interradial marginal plates and 3 (*O. crinita*) or 3–4 (*O. anisacantha*) arm spines, but both with dorsal arm plates present (Clark 1911; Franklin and O’Hara 2008).



FIGURE 5. Limestone fragments collected from the rock dredge sampling station 18 during the INTEMARES-A22B_0718 survey. The holotype of *Ophiomyxa xispa* sp. nov. was collected from a crevice in the largest fragment.

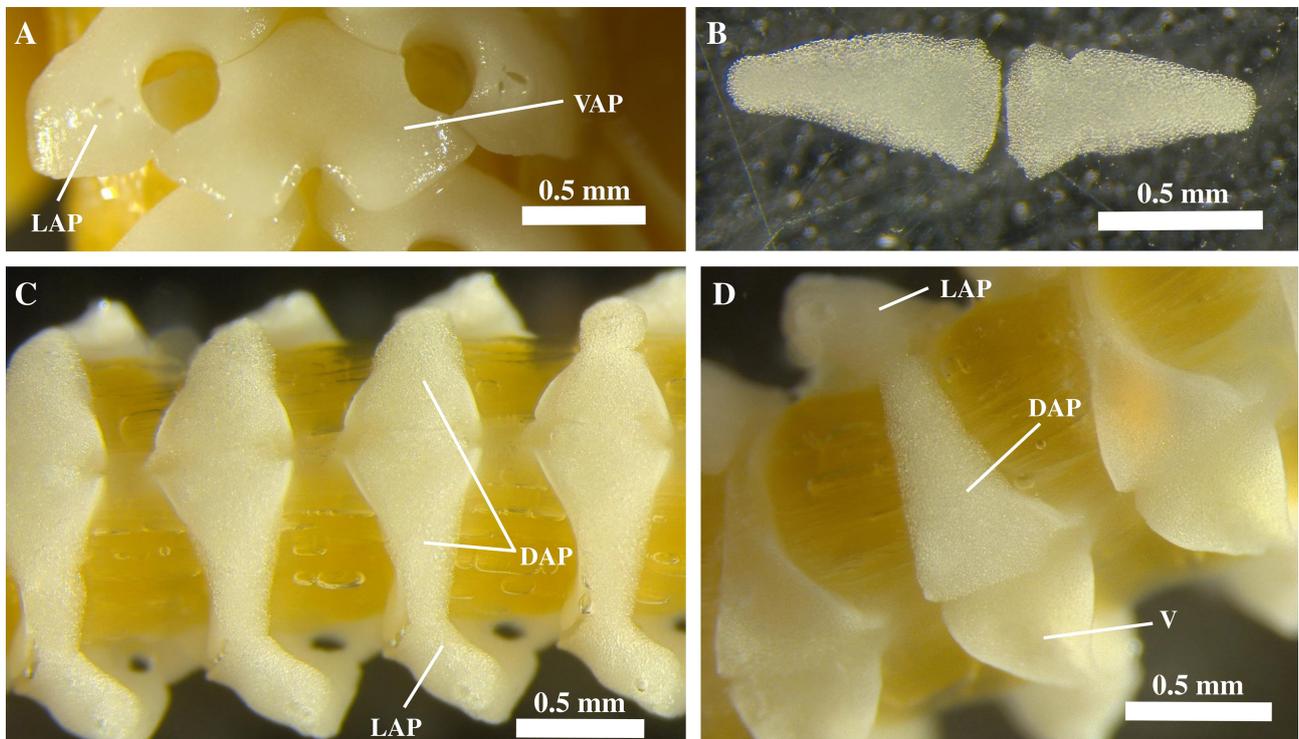


FIGURE 6. *Ophiomyxa pentagona*, individual CBB-UIB 100487 after treatment in 4% sodium hypochlorite. A: ventral side of the arm showing a proximal ventral arm plate (VAP). B: two dorsal arm plates (DAP) removed from the arm. C: dorsal side of arm showing the DAPs covering the vertebrae. D: dorsal side of arm showing on one side the nude vertebra (V), the other side still covered by the DAP. LAP, lateral arm plate.

Within *Ophiodera* and related species, our species morphologically resembles the most to *O. anisacantha*, *O. bengalensis*, *O. crinita*, *O. neglecta* and *O. serpentaria*, due to the combination of the absence of interradial marginal plates and the presence of three arm spines. However, it is easily distinguished by the shape and number of dorsal arm plates. Dorsal arm plates are small, fragmented (3–4 fragments) and only present in the proximal of the arm in *O. bengalensis* (Mortensen 1933a), and also presents the disk covered by rounded granulations (Koehler 1897). *Ophiomyxa serpentaria* was described as lacking dorsal arm plates (Lyman 1883; Mortensen 1927; Paterson 1985), but Mortensen (1933b) re-described this species, reporting the presence of one thin and transparent, regularly perforated large dorsal arm plate with semicircular shape in each segment, the rounded side oriented distally. This dorsal arm plates could also be observed in the sequenced individuals DZMB-HH 61037 (GenBank ID: ON341503) and DZMB-HH 61728 (GenBank ID: ON341511). Furthermore, *O. serpentaria* has ventral arm plates widely in contact (Mortensen 1927) vs. separated in our species. *Ophiomyxa anisacantha* has two larger dorsal arm plates on each segment, one on each side of the midline of the arm, where they are in touch, and a variable number of smaller plates proximally, connecting with the larger plates of the previous segment (Clark 1911). *Ophiomyxa crinita* has a large rhombic dorsal arm plate accompanied by three smaller ones, one overlaying the proximal edge of the large one and two proximal, one on each side of the arm midline (Franklin and O'Hara 2008). *Ophiomyxa neglecta* has a pair of large dorsal arm plates, one on each side of the arm midline which are not in contact but the space between them is filled by smaller plates (Koehler 1922). Moreover, among all these species, the presence of ossicles has only been reported for three of them, *O. crinita* and *O. neglecta*, both with C-shaped ossicles, in the stomach wall, and in the disk and arms integument, respectively, and *O. serpentaria*, with bone-shaped ossicles in the disk integument (Mortensen 1933b). We have not detected this type of ossicle in our species, but instead we record the presence of perforated plates in the disk integument, similar to those present in some holothurians.

The phylogenetic clade in which *O. xispa* **sp. nov.** is included in one group gathering species with an Indian Ocean (*O. bengalensis* and *O. crinita*) or western Pacific Ocean distribution (*O. neglecta*) (Koehler 1897, 1904; Franklin & O'Hara 2008), and another group including *O. stimpsonii*, distributed in the Caribbean Sea (western Atlantic Ocean) (Verrill 1899), and *O. xispa* **sp. nov.**, so far only known from the Mediterranean. Paradoxically, the geographically closer species *O. serpentaria* and *O. pentagona*, distributed in the north eastern Atlantic Ocean and north eastern Atlantic Ocean and Mediterranean Sea, respectively (Mortensen 1927; Tortonese 1965), belong to the other detected phylogenetic clade, which also includes species distributed in the Caribbean (*O. tumida*, *O. brevicauda*, *O. flaccida*, the latter also southwards at least to Brazil) (Say 1825; Lyman 1883; Verrill 1899; Gondim *et al.* 2013), the western Pacific Ocean (*O. australis*, *O. brevissima*, *O. anisacantha*) and the south Atlantic Ocean (*O. vivipara*) (Studer 1876; Mortensen 1936). Besides the widespread distribution of the representatives of the two clades, so far, we have not been able to detect distinctive consistent morphological traits allowing to discern among them.

Discussion

In the Mediterranean, the scientific knowledge about seamounts contains large gaps, mainly on the eastern basin, but also by an asymmetry between the amount of geological and biological studies (Würtz *et al.* 2015). Recent surveys carried out in the Mallorca Channel seamounts, including Ausias March, Ses Olives and Emile Baudot seamounts (Massutí *et al.* 2022), have substantially improved our knowledge of Mediterranean invertebrates. This improvement has been particularly important for sponges, with the description, so far, of seven new species (Díaz *et al.* 2021; 2023a), but also echinoderms, with the report of the first Mediterranean record of a brittle star of the family Ophiolidae Perrier, 1873, *Ophiomyces grandis* Lyman, 1879, which showed high abundances in these seamounts despite its absence from insular coasts (Ordines *et al.* 2019).

Our findings emphasize the role played by seamounts as specialised rocky habitats that support a fauna that does not appear to occur along continental margins. The first studies of Mediterranean seamounts considered these geomorphological structures as islands, which may serve as isolated refuges for relict populations of species from a previously larger distribution (Galil & Zibrowius 1998). However, it is also possible that specialized seamount communities exist that are connected through dispersal, as suggested by Clark *et al.* (2010; and references therein). Whatever the actual situation or even if they occur together, seamounts appear to represent a unique habitat necessary for some species that apparently cannot establish permanent populations along continental margins, even when they are close. This is the case of Mallorca Channel seamounts fauna in which, despite being quite close to the insular

coasts (only just 10–15 nm apart), most of the new species or new Mediterranean records are not known from anywhere else in the Mediterranean. Five out of the seven new sponge species recently described from the Mallorca Channel seamounts have only been collected there, despite thorough benthos sampling efforts that covered the Balearic Islands and adjacent Iberian Peninsula coast in the last years (Díaz *et al.* 2021; 2023a,b). The same situation exists for *O. grandis*, an ophiuroid whose reported distribution is almost exclusively associated to isolated islands or seamounts (Ordines *et al.* 2019). Although so far, we only have one specimen of *O. xispa* **sp. nov.** and further records could indicate a wider habitat preference, it is remarkable that it was collected from the Mallorca Channel seamounts, and hence represents one more reason to prioritize their protection as unique habitats that sustain rare species populations.

Acknowledgements

The authors would like to thank very much Sabine Stöhr for her kind help providing some of the original descriptions listed in the bibliographic references. We also thank Dr. Tim O'Hara (Museums Victoria) for kindly providing us COI sequences belonging to the species *O. brevicauda*, *O. stimpsonii* and *O. tumida* as well as revisions made to this study. These three specimens were identified and sequenced from the collections of the Muséum National D'Histoire Naturelle (MNHN, Paris, France) from their 2015 KARUBENTHOS campaign to Guadeloupe. We also thank Dr. Saskia Brix-Elsig and Dr. Antje Fischer for kindly providing us with *O. serpentaria* specimens from the Deutsches Zentrum für Marine Biodiversitätsforschung (DZMB) collection. We also thank Dr. Ana Ramos (Centro Oceanográfico de Vigo) and Dr. Fran Ramil (Universidade de Vigo) for kindly for letting us review the material belonging to *O. serpentaria*. We thank all participants who took part in the research surveys INTEMARES_A22B_0718, INTEMARES_A22B_1019, INTEMARES_A22B_0720, INTEMARES_A22B_0820, as well as the captains and crew of the R/Vs *Ángeles Alvariño* and *Sarmiento de Gamboa*.

This research has been performed in the scope of the LIFE IP INTEMARES research project “Gestión integrada, innovadora y participativa de la Red Natura 2000 en el medio marino español”, coordinated by the Biodiversity Foundation of the Spanish Ministry for the Ecological Transition and the Demographic Challenge and co-funded by the European Union’s LIFE program (LIFE15 IPE ES012). Financial support has also been received from the 18-ESMARES2-CIRCA research project, included in the scientific program “Asesoramiento científico-técnico para la protección del medio marino: Evaluación y seguimiento de las Estrategias Marinas, Seguimiento de los espacios marinos protegidos de competencia estatal (2018–2021)”, co-funded by the General Directorate of Sea Protection of the Spanish Ministry for the Ecological Transition and the Demographic Challenge, as well as from the research project TAXOMAB “Una aproximación taxonómica integrativa para mejorar el conocimiento de la biodiversidad marina del Archipiélago Balear” co-funded by Regional Government of the Balearic Islands (BIO023) and framed within the Complementary Plans Recovery, Transformation and Resilience Plan. S. Ramírez-Amaro is supported by a postdoctoral contract co-funded by the Regional Government of the Balearic Islands and the European Social Fund.

References

- Anadón, R. (1977) Equinodermos recogidos durante la campaña “Atlor VII” en las costas noroccidentales de África (noviembre 1975). *Resultados Expediciones Científicas del Buque Oceanográfico Cornide de Saavedra*, 6, 165–168.
- Clark, M.R., Rowden, A.A., Schlacher, T., Williams, A., Consalvey, M., Stocks, K.I., Rogers, A.D., O'Hara, T.D., White, M., Shank, T.M. & Hall-Spencer, J.M. (2010) The ecology of seamounts: structure, function, and human impacts. *Annual Reviews in Marine Science*, 2, 253–278.
<https://doi.org/10.1146/annurev-marine-120308-081109>
- Clark, A.H. (1952) Echinoderms from the Marshall Islands. *Proceedings of the U.S. National Museum*, 102, 265–303.
<https://doi.org/10.5479/si.00963801.102-3302.265>
- Clark, H.L. (1911) North Pacific Ophiurans in the collection of the United States National Museum. *Smithsonian Institution United States National Museum Bulletin*, 75, 1–302.
<https://doi.org/10.5962/bhl.title.32556>
- Corstorphine, E.A. (2010) *DNA barcoding of echinoderms: Species diversity and patterns of molecular evolution*. PhD Thesis, University of Guelph, Guelph, 144 pp.
- Díaz, J.A., Ramírez-Amaro, S. & Ordines, F. (2021) Sponges of Western Mediterranean seamounts: new genera, new species and new records. *PeerJ*, 9, e11879.

<https://doi.org/10.7717/peerj.11879>

- Díaz, J.A., Ordines, F., Massutí, E. & Cárdenas, P. (2023a) From caves to seamounts: the hidden diversity of tetractinellid sponges from the Balearic Islands, with the description of eight new species. *PeerJ*. [published online]
- Díaz, J.A., Ordinas, F., Fariols, M.T., Melo-Aguilar, C. & Massutí, E. (2023b) Sponge assemblages in fishing grounds and seamounts of the Balearic Islands (western Mediterranean). *Deep-Sea Research*, Part I, 104211.
<https://doi.org/10.1016/j.dsr.2023.104211>
- Díaz, J.A., Ordines, F., Massutí, E. & Cárdenas, P. (2023) From caves to seamounts: the hidden diversity of tetractinellid sponges from the Balearic Islands, with the description of eight new species. *PeerJ*. [in press]
- Eichsteller, A., Taylor, J., Stöhr, S., Brix, S. & Martínez Arbizu, P. (2022) DNA Barcoding of Cold-Water Coral-Associated Ophiuroid Fauna from the North Atlantic. *Diversity*, 14, 358.
<https://doi.org/10.3390/d14050358>
- Folmer, O., Black, M., Hoeh, W., Lutz, R. & Vrijenhoek, R. (1994) DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology*, 3, 294–299.
- Franklin, A. & O'Hara, T. (2008) A new species in the genus *Ophiomyxa* from South-west Australian waters (Echinodermata: Ophiuroidea: Ophiomyxidae). *Memoirs of Museum Victoria*, 65, 57–62.
<https://doi.org/10.24199/j.mmv.2008.65.5>
- Galil, B. & Zibrowius, H. (1998) First benthos samples from Erastothernes Seamount, eastern Mediterranean. *Senckenbergiana Maritima*, 28 (4/6), 111–121.
<https://doi.org/10.1007/BF03043142>
- Gondim, A.I., Alonso, C., Dias, T.L.P., Manso, C.L.C. & Christoffersen, M.L. (2013) A taxonomic guide to the brittle-stars (Echinodermata, Ophiuroidea) from the State of Paraíba continental shelf, Northeastern Brazil. *ZooKeys*, 307, 4596.
<https://doi.org/10.3897/zookeys.307.4673>
- Hall, T.A. (1999) BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acids Symposium Series*, 41, 95–98.
- Hendler, G. (2018) Armed to the teeth: a new paradigm for the buccal skeleton of brittle stars (Echinodermata: Ophiuroidea). *Contributions in Science*, 526, 189–311.
<https://doi.org/10.5962/p.324539>
- Hugall, A.F., O'Hara, T.D., Hunjan, S., Nilsen, R. & Moussalli, A. (2016) An Exon-Capture System for the Entire Class Ophiuroidea. *Molecular Biology and Evolution*, 33 (1), 281–294.
<https://doi.org/10.1093/molbev/msv216>
- Koehler, R. (1897) Echinodermes recueillis par "l'Investigateur" dans l'Océan Indien. I. Les Ophiures de mer profonde. *Annales des Sciences Naturelles Zoologie*, 8, 277–372.
- Koehler, R. (1922) Ophiurans of the Philippine Seas and adjacent waters. Contributions to the biology of the Philippine Archipelago and adjacent regions. *Smithsonian Institution United States National Museum Bulletin*, 100, 1–624.
<https://doi.org/10.5962/bhl.title.32917>
- Lyman, T. (1875) Zoological Results of the Hassler Expedition. 2. Ophiuridae and Astrophytidae. *Illustrated catalogue of the Museum of Comparative Zoology at Harvard College*, 8 (2), 1–34.
- Lyman, T. (1883) Reports on the results of dredging, under the supervision of Alexander Agassiz, in the Caribbean Sea (1878–79), and on the east coast of the United States, during the summer of 1880, by the U.S. coast survey steamer "Blake", commander J.R. Bartlett, U.S.N., commanding. XX. Report on the Ophiuroidea. *Bulletin of the Museum of Comparative Zoology at Harvard*, 10 (6), 227–287.
- Massutí, E., Sánchez-Guillamón, O., Fariols, M.T., Palomino, D., Frank, A., Bárcenas, P., Rincón, B., Martínez-Carreño, N., Keller, S., López-Rodríguez, C., Díaz, J.A., López-González, N., Marco-Herrero, E., Fernandez-Arcaya, U., Valls, M., Ramírez-Amaro, S., Ferragut, F., Joher, S., Ordinas, F. & Vázquez, J-T. (2022) Improving Scientific Knowledge of Mallorca Channel Seamounts (Western Mediterranean) within the Framework of Natura 2000 Network. *Diversity*, 14 (1), 4.
<https://doi.org/10.3390/d14010004>
- Matsumoto, H. (1915) A new classification of the Ophiuroidea: with descriptions of new genera and species. *Proceedings of the Academy of Natural Sciences of Philadelphia*, 67, 43–92.
- Mortensen, T. (1927) *Handbook of the Echinoderms of the British Isles*. Oxford University Press, Oxford, 471 pp.
<https://doi.org/10.5962/bhl.title.6841>
- Mortensen, T. (1933a) Papers from Dr. Th. Mortensen's Pacific Expedition 1914–16. LXV. Echinoderms of South Africa. *Videnskabelige Meddelelser fra Dansk Naturhistorisk Forening*, 93, 215400.
- Mortensen, T. (1933b) Ophiuroidea. *Danish Ingolf Expedition*, 4 (8), 1–121.
- Mortensen, T. (1936) Echinoidea and Ophiuroidea. *Discovery Reports*, 12, 199348.
<https://doi.org/10.5962/bhl.part.8051>
- O'Hara, T.D., Stöhr, S., Hugall, A.F., Thuy, B. & Martynov, A. (2018) Morphological diagnoses of higher taxa in Ophiuroidea (Echinodermata) in support of a new classification. *European Journal of Taxonomy*, 416, 1–35.
<https://doi.org/10.5852/ejt.2018.416>
- Okanishi, M. & Fujita, T. (2013) Molecular phylogeny based on increased number of species and genes revealed more robust family-level systematics of the order Euryalida (Echinodermata: Ophiuroidea). *Molecular Phylogenetic and Evolution*, 69 (3), 566–580.

<https://doi.org/10.1016/j.ympcv.2013.07.021>

- Okanishi, M. & Fujita, Y. (2018) A new species of *Ophioconis* (Echinodermata: Ophiuroidea) from Ryukyu Islands, southwestern Japan. *Proceedings of the Biological Society of Washington*, 131 (1), 163–174.
<https://doi.org/10.2988/18-00001>
- Ordines, F., Ramírez-Amaro, S., Fernández-Arcaya, U., Marco-Herrero, E. & Massutí, E. (2019) First occurrence of an Ophiohelidae species in the Mediterranean: the high abundances of *Ophiomyces grandis* from the Mallorca Channel seamounts. *Journal of the Marine Biological Association of the United Kingdom*, 99 (8), 1817–1823.
<https://doi.org/10.1017/S0025315419000808>
- Paterson, G.L.J. (1985) The deep-sea Ophiuroidea of the North Atlantic Ocean. *Bulletin of the British Museum (Natural History)*, 49, 1–162.
- Reiss, H., Kröncke, I. & Ehrich, S. (2006) Estimating the catching efficiency of a 2-m beam trawl for sampling epifauna by removal experiments. *ICES Journal of Marine Science*, 63, 1453–1464.
<https://doi.org/10.1016/j.icesjms.2006.06.001>
- Ronquist, F., Teslenko, M., Van Der Mark, P., Ayres, D.L., Darling, A., Höhna, S. & Huelsenbeck, J.P. (2012) MrBayes 3.2: Efficient Bayesian phylogenetic inference and model choice across a large model space. *Systematic Biology*, 61 (3), 539–542.
<https://doi.org/10.1093/sysbio/sys029>
- Say, T. (1825) On the species of the Linnaean genus *Asterias* inhabiting the coast of the U.S. *Journal of the Academy of natural sciences of Philadelphia*, 5, 141–154.
- Smith, A.B., Paterson, G.L.J. & Lafay, B. (1995) Ophiuroid phylogeny and higher taxonomy: morphological, molecular and palaeontological perspectives. *Zoological Journal of the Linnean Society*, 114, 213–243.
<https://doi.org/10.1006/zjls.1995.0024>
- Stöhr, S., O’Hara, T.D. & Thuy, B. (2012) Global diversity of brittle stars (Echinodermata: Ophiuroidea). *PLoS ONE*, 7, e31940.
<https://doi.org/10.1371/journal.pone.0031940>
- Stöhr, S., O’Hara, T.D. & Thuy, B. (Eds.) (2024) World Ophiuroidea Database. Available from: <https://www.marinespecies.org/ophiuroidea> (accessed 22 January 2024)
- Tamura, K., Stecher, G., Peterson, D., Filipowski, A. & Kumar, S. (2013) MEGA6: Molecular Evolutionary Genetics Analysis Version 6.0. *Molecular Biology and Evolution*, 30, 2725–2729.
<https://doi.org/10.1093/molbev/mst197>
- Thompson, J.D., Higgins, D.G. & Gibson, T.J. (1994) CLUSTAL W: Improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. *Nucleic Acids Research*, 22 (22), 4673–4680.
<https://doi.org/10.1093/nar/22.22.4673>
- Tortonese, E. (1965) *Fauna d’Italia. Echinodermata*. Edizioni Calderini, Bologna, 419 pp.
- Verrill, A.E. (1899) Report on the Ophiuroidea collected by the Bahama expedition in 1893. *Bulletin of the Laboratories of Natural History of the State of Iowa*, 5, 1–88.
- Verrill, A.E. (1900) VII. North American Ophiuroidea. I.-Revision of certain families and genera of West Indian Ophiurans. II.-A faunal catalogue of the known species of West Indian Ophiurans. *Transactions of the Connecticut Academy of Arts and Sciences*, 10, 301–371.
<https://doi.org/10.5962/bhl.part.7032>
- Würtz, M., Rovere, M. & Bo, M. (2015) Introducing the Mediterranean seamount atlas: general aspects. In: Würtz, M. & Rovere, M. (Eds.), *Atlas of the Mediterranean Seamounts and Seamount-Like Structures*. IUCN, Málaga, pp. 11–19.
<https://doi.org/10.2305/IUCN.CH.2015.07.en>